Analyzing Blood Lead Specimens at the Kansas Department of Health and Environmental Laboratories

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KDHE Laboratory Lead History

- Early 1990's GFAA, limited to whole blood analysis submitted by County Health Departments
- Mid 1990's advances in GFAA made it possible to increase analysis, capillary or venous whole blood samples (4000 samples/year)
- Late 1990's , KDHE Laboratory allowed other facilities to submit samples, however screening remained relatively flat
- By 2000 ICP-MS emerged into clinical analysis
 - ICP-MS widely used for environmental analysis
- 2001 KDHE Laboratory purchased a second ICP-MS with intent on shifting all GFAA analysis to ICP-MS
- Whole blood Screening continued to decline (2500 sample/year)

ICP-MS

- Inductively coupled argon plasma mass spectroscopy
- Energy of Plasma breaks down a sample into its elemental state
- Separate metals based on mass
- Lead (Pb) has 3 major IsotopesPb 206, Pb 207, and Pb 208
- ICP-MS allows much lower detection limits
 - Multiple analytes
 - Faster
- KDHE completed transition of all analysis from GFAA to ICP-MS

What Does This Have To Do With You???

- ICPMS allows the laboratory to analyze blood leads using different sample collection techniques.
- Two ways to collect a blood lead.
 - Whole blood venous draw in an EDTA tube
 - Capillary finger stick onto a filter paper



Sample Type

- Whole blood/Venous draw is considered a diagnostic test which can be used for initial blood lead level or confirmation of a screening sample.
- Capillary/finger stick is considered a screening test defined as any capillary draw either whole blood collected in microtainers or whole collected on filter paper.
- With that said why collect a capillary sample on filter paper?

Advantages of Filter Paper

- Allows increased screening for blood lead in children
- Less traumatic and easier to collect
- Specimen transportation
- Cost effective

Disadvantages of Filter Paper

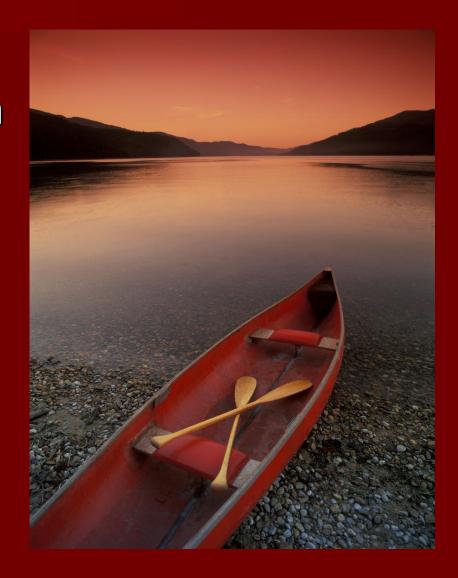
- Contamination
- Filter paper background
- Small sample size
- Accuracy
- Extended prep time

Early Work

- Early work had been done by Demshar and Wang for filter paper analysis using GFAA
- KDHE Goal-Increase lead screening within State
 - Clinicians asking about filter paper screening
 - No methodology was available for ICP-MS
 - Controversial
 - Only one Proficiency program with 4 participants
 - No quality control samples available
- KDHE Laboratory asked to develop method to analyze dried blood spots for lead
- And So The Journey Begins...

The Journey of a Blood Spot

- Filter paper Collection
- Shipping & Receiving
- Sample Preparation
- Analysis
- Reporting
- Follow-up



Blood Lead Order Form

- Blood Lead forms can be ordered from the KDHE Requisition Form
- http://www.kdheks.gov/l abs/packaging_and_ship ping.html
- Must have Facility ID to order
- Order forms, lancets, confirmation kits (EDTA)



MR-1 Rev. 11/21/05

DIVISION OF HEALTH AND ENVIRONMENTAL LABORATORIES DEPARTMENT OF HEALTH AND ENVIRONMENT

Forbes Field, Building 740 Topeka, Kansas 66620-0001

REQUISITION FOR LABORATORY SPECIMEN KITS

Please use the appropriate kit listed below to submit specimens to the Health and Environmental Laboratories. Each kit consists of a specimen container, an addressed mailing container, and a kit requisition form. Order the Universal Specimen Submission forms in the space below. If you have any questions about submitting specimens, please refer to the Manual of Laboratory Tests or call (785) 296-1623. Please enter the quantity needed on the line next to the item.

RUSH ORDERS: FAX to (785) 296-1641 Universal Specimen Submission Forms Only Specify number required Blood Lead Filter Paper Forms Blood Lead Confirmation Kits EDTA (Purple Top) Blood Tubes Multi-tube bottle with mailing box (5 tube box) Blood Tubes (Yellow Top) Chlamydia/Gonorrhea Mailer Initial (Green) Collection Unit - 🗆 Eng 🔲 Span Repeat (Red) Collection Unit - 🗖 Eng 🗖 Span Viral Culture Virus VTM (HD and Influenza Flu VTM Surveillance Sites Only) Miscellaneous Infectious Disease (IDS) Shipper Parasite (O & P) Feces Mailer Pinworm (Health Departments Only) Sputum Mailer Culture Plates Pertussis Mailer Mailer, for two specimens CO2 Tablets Whirl-Pak Bag (Specify): Contact Epidemiologic Services at (877) 427-7317 first for AIDS C/T test, Prenatal tests and WNV tests Facility ID No. : LAB USE ONLY Order Number: _ Date Received: Date Shipped: _ Shipped By:

Filter Paper Collection

- Fill Out Form Completely
- Include Medicaid Number if Applicable
- Mark Correct Collection Type
- Read Directions on Back of Form and Coverlet Prior to Collection
- Collect Blood Spots on Filter Paper
- Allow Blood Spots to Dry

Blood Lead Collection Form



Collecting the Blood Spots









Blood Spot Collection



- During collection the blood spots should never come in contact with the coverlet or any other surface.
- Allow the blood spots to dry a minimum of 2 hours.

Shipping the Specimen

- After the blood spots are dry, fold the coverlet over the blood spots to act as a primary cover.
- Blood Lead Specimens can be shipped in a standard envelop.
- Use the label on the form to address the envelop.





Receiving the Specimen

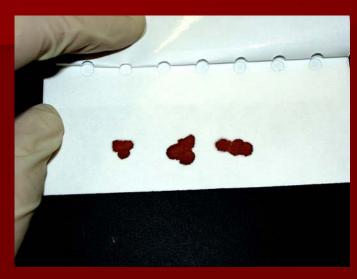
- Blood Lead Specimens are received at the laboratories via USPS, UPS, FedEx, and courier.
- Blood Lead Forms are reviewed for missing information.
- Blood Specimens are evaluated for quality and flagged if unsatisfactory.

The Good, The Bad & The Ugly Satisfactory Spots





The Good, The Bad & the Ugly Unsatisfactory Spots









Sample Preparation

- Punch a ¼ inch blood spot into a sample tube.
- Addition of water/Triton X to 1/4 inch blood spot. Mix.
- Addition of nitric acid and Tb internal standard. Mix.





Sample Preparation

- Centrifuge specimen.
- Filter sample into new sample tube avoiding filter paper particulates.
- Direct aspiration of sample into the ICPMS plasma.





Analysis Dilution Factor with Aqueous Standards



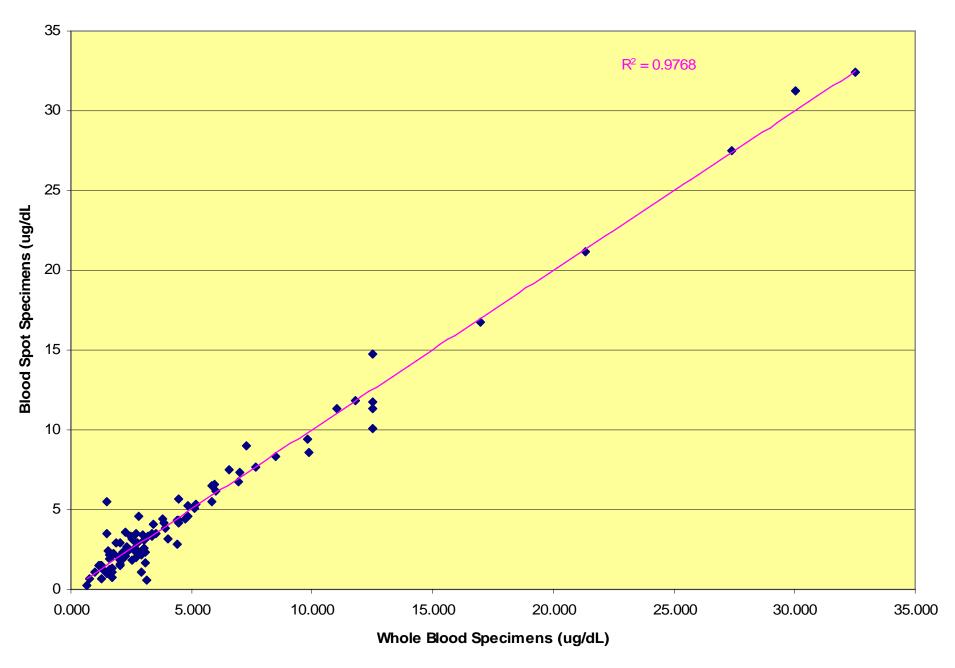


- Demshar & Wang (Analyst June 1992 Vol 117)
- 6.7 µL Whole Blood per 3/16 inch punch
- KDHE Calculated (Based on Publication)
- 11.9 µL Whole Blood per 1/4 inch punch

Dilution Factor

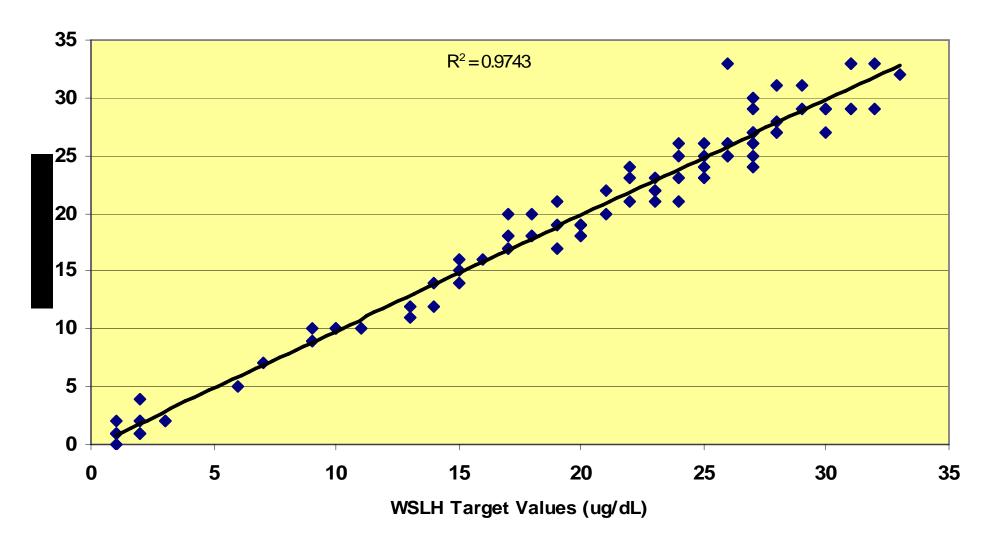
- Theoretically 11.9 μL Extracted into 500 μL of Diluent gives a Dilution Factor of 42
- KDHE Experimental Data Based on Proficiency and Patient samples results in a Dilution Factor of 45

Correlation Between Whole Blood and Blood Spots



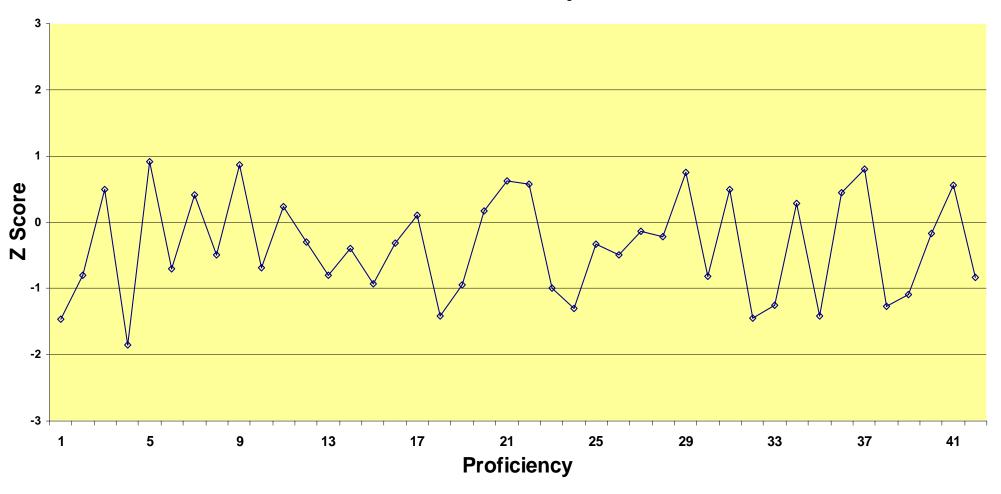
Original data. Patient whole blood analyzed by ICP-MS and then spotted on filter paper. Blood spot analyzed by same ICP-MS. (n=100)

Filter Paper Proficiency Test Values



Monthly Wisconsin State Laboratory of Hygiene Proficiency values from March 2003 – August 2005 (n=100)

Z Scores for 2006 Blood Spot Proficiencies



Analysis Continued...

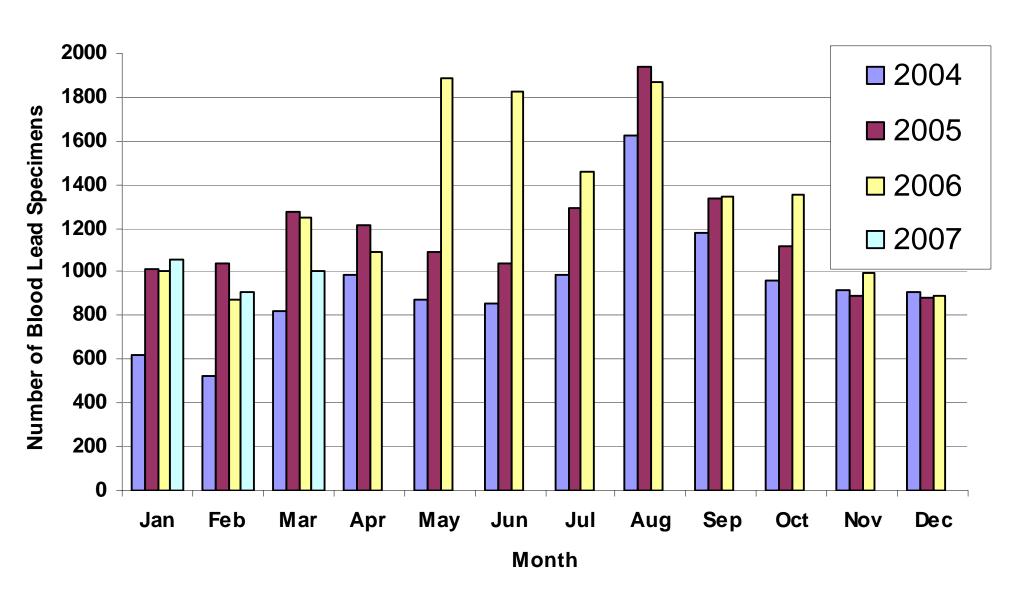
- Every sample submitted to the Laboratory will be analyzed.
- Every filter paper blood lead result > 14 ug/dL will be reanalyzed for confirmation.



Reporting Results & Follow-up

- Blood Lead results are mailed to the submitter and electronically sent to the CLPPP.
- A comment will be added to any report with a lead value > 20 ug/dL.
 - A venous draw is recommended for every blood lead confirmation.
- The Laboratory will fax all results ≥ 20 ug/dL.
- The Laboratory will fax and call the submitter on any results > 40 ug/dL.

Month by Month Comparison



Elevated Blood Leads Are Still a Concern (Numbers based on 2006 data)

■ 15,848 blood leads analyzed in 2006

■ 5-10 ug/dL: 1,374

■ 10-20 ug/dL: 289

■ 20-44 ug/dL: 70

■ >44 ug/dL: 6

Conclusion

- ICP-MS provides low detection limits and allows the option of multiple elemental analyses on the same matrix at the same time.
- ICP-MS is an excellent resource for the detection of lead in whole blood.
- ICP-MS can effectively be used as a screening program for blood lead using filter paper spots.
- Filter paper collection increased the number of patients screened for blood lead in the state of Kansas by 540%.

